

P10455-IM

Immortalized Human Cardiac Microvascular Endothelial Cells have been established by transducing primary human cardiac microvascular endothelial cells with the SV40 large T antigen (SV40T), enabling prolonged proliferation while preserving key endothelial functions. These cells form the inner lining of the cardiac microvasculature and are critical for regulating vascular tone, myocardial perfusion, barrier integrity, and inflammatory signaling.

Cardiac microvascular endothelial dysfunction contributes to pathologies such as heart failure with preserved ejection fraction (HFpEF), microvascular angina, and ischemia-reperfusion injury. The immortalized cells retain expression of characteristic endothelial markers and secrete functional mediators including VEGF, ICAM-1, and nitric oxide, making them a robust and reproducible model for cardiovascular research, disease modeling, and pharmacological testing.



IMMORTALIZED HUMAN CARDIAC MICROVASCULAR ENDOTHELIAL CELLS

Product Type: Immortalized Human Cardiac Microvascular Endothelial Cells

Catalog Number: P10455-IM

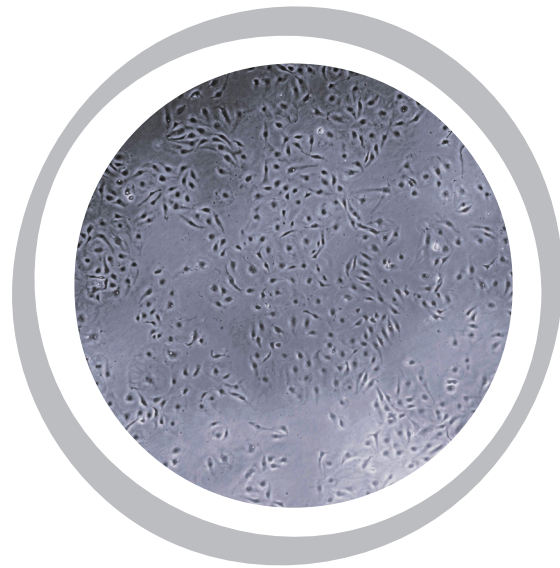
Immortalization: SV40T. G418 resistant.

Number of cells: >1x10⁶ cells (cryopreserved vials)

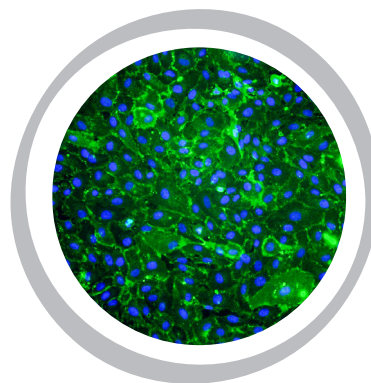
Storage: Liquid Nitrogen

Recommended Medium: Endothelial Cell Medium Kit (Ref: P60104)

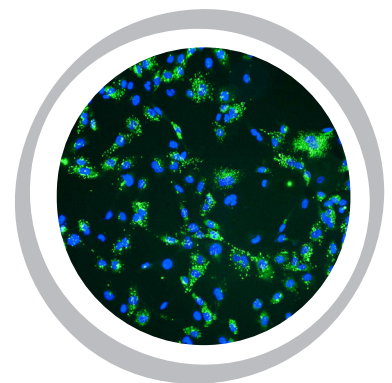
Product Characterization: Positive for vWF/Factor VIII, CD31 (P-CAM) and tubule formation.



Phase-contrast



CD31



vWF

THIS PRODUCT IS FOR RESEARCH PURPOSES ONLY

It is not to be used for drug or diagnostic purposes, nor is it intended for human use. Innoprot products may not be resold, modified for resale, or used to manufacture commercial products without written approval of Innovative Technologies in Biological Systems, S.L.

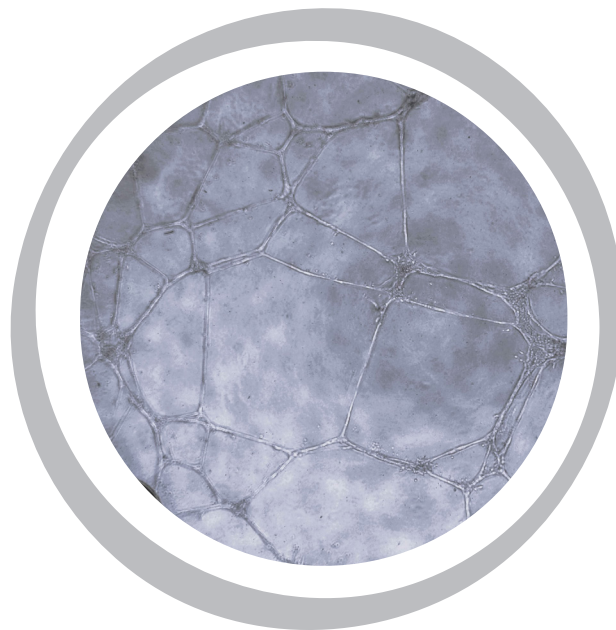
About Immortalized Human Retinal Endothelial Cells

The immortalized human cardiac microvascular endothelial cell line has been established through genetic modification of primary human cardiac microvascular endothelial cells using the SV40 large T antigen (SV40T) as the sole immortalizing agent. SV40T, derived from the Simian Virus 40 Large T-antigen, interferes with key regulators of the cell cycle, allowing the cells to bypass replicative senescence and achieve extended proliferative capacity.

This single-factor approach enables the generation of a stable cell line with long-term growth potential, suitable for repeated passaging and consistent experimental use. While primary human cardiac microvascular endothelial cells typically lose proliferative capacity after approximately five passages, SV40T-transduced cells maintain viability and expansion potential beyond 20 passages. This makes them a reliable tool for in vitro studies focused on cardiovascular endothelial biology, microvascular dysfunction, and therapeutic screening.

Functional Assay: Tube Formation

To evaluate the angiogenic functionality of the immortalized cell line, a tube formation assay was performed. Cells were seeded into Geltrex-coated 96-well plates at a density of 18×10^3 cells per well. This extracellular matrix surrogate supports the organization of endothelial cells into capillary-like structures. After 24 hours of incubation at 37°C in a humidified atmosphere with 5% CO₂, the tube formation was verified.



Tubule formation

Culturing conditions

1 IMMEDIATELY UPON DELIVERY

- 1.1 Remove the vial from the shipping container to check for freezing.
- 1.2 Transfer the frozen vial to liquid nitrogen until ready to thaw.

2 THAWING CELLS:

- 2.1 Fibronectin coating step (optional): Prepare a fibronectin coated flask (2 µg/cm²) is recommended depending on the type of flasks you are using (i.e. if not using pre-treated flasks for endothelial cells). Add 3 ml of sterile Dulbecco's phosphate buffered saline (DPBS) to a T-25 flask and then add 50 µl of fibronectin stock solution (1 mg/ml, Innoprot cat. no. P8248). Leave the flask in incubator overnight. Aspirate fibronectin solution and add 15 ml of complete medium to the culture vessel. The fibronectin solution can be used twice. Leave the vessel in the sterile field and proceed to thaw the cryopreserved cells.
- 2.2 Prepare "Thawing medium" by combining 500 ml of basal medium, 25 ml of fetal bovine serum, 5 ml of Growth supplement and 5 ml of penicillin/streptomycin solution.
- 2.3 Thaw cells rapidly in a 37°C water bath; avoid allowing the sample to warm to 37°C. Cryovials should be cool to the touch when removed.
- 2.4 Remove the vial, wipe it dry, and transfer it to a sterile field.
- 2.5 Rinse the vial with 70% ethanol, then wipe to remove excess. Open the vial and resuspend its contents using a 1 ml Eppendorf pipette.
- 2.6 Dispense the contents into a 25 cm² culture flask with warm complete media (FBS percentage can be increased up to 10% for better culture establishment).
- 2.7 Place the flask in the incubator.
- 2.8 For optimal results, avoid disturbing the culture for 16 hours after initiation. Change the growth medium the next day to remove unattached cells, then every other day thereafter.

3 MAINTENANCE OF THE CULTURE:

- 3.1 Change medium 48 hours after establishing a subculture.
- 3.2 Subculture when cells are over 90% confluent.

4 SUBCULTURING:

Remove medium, rinse with 0.05% trypsin-EDTA solution. Add 1 to 2 mL of trypsin-EDTA solution and allow the flask to sit until cells detach. Add fresh culture medium, aspirate, and dispense into new culture flasks. Recommended subcultivation ratio of 1:2 to 1:3. Medium Renewal: 2 to 3 times per week.

Reagents for cryopreservation: Cryostor S10.

Quality Control / Biosafety

The cells test negative for HIV-1, HBV, HCV, mycoplasma, bacteria, yeast and fungi.